

Optimum Sample Concentration for Resonance Raman Spectra

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In resonance Raman spectroscopy, absorption of incident radiation and of scattered radiation increases with sample concentration. Scattering intensity at the focal point of the laser also increases with sample concentration. An optimum sample concentration is sought where the scattered radiation intensity emerging from the sample cell is at a maximum. Assuming a point-scattering model, Strekas *et al.*¹ have derived the relationship:

$$I = I_0 J c e^{-k(r\epsilon_s + l\epsilon_i)} \quad (1)$$

I is the observed intensity, I_0 the laser intensity on entry into the sample cell, J is the molar scattering coefficient, c is the molar concentration, ϵ_s and ϵ_i are the decadic molar absorptivities at scattering wavelength and the incident wavelength, r and l are the pathlengths of the laser beam and the scattered radiation in the Raman cell, respectively, and $k = \ln 10 = 2.303$. This function has a maximum. Setting the differential with respect to concentration equal to zero gives the optimum concentration:

$$c_{\text{opt}} = [k(r\epsilon_s + l\epsilon_i)]^{-1} \quad (2)$$

If $r = l$, Eq. (2) reduces to:

$$c_{\text{opt}} = [kr(\epsilon_s + \epsilon_i)]^{-1} \quad (3)$$

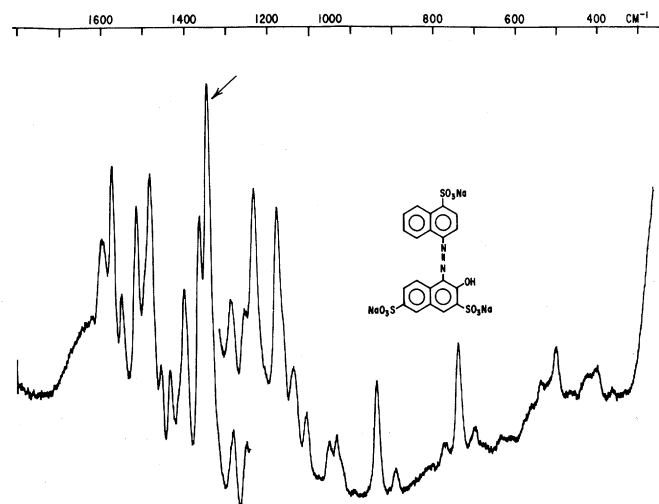


FIG. 1. Resonance Raman spectrum of amaranth, 488.0 nm excitation, laser power ~ 300 mW at the sample. Line indicated with arrow was used for intensity measurements.

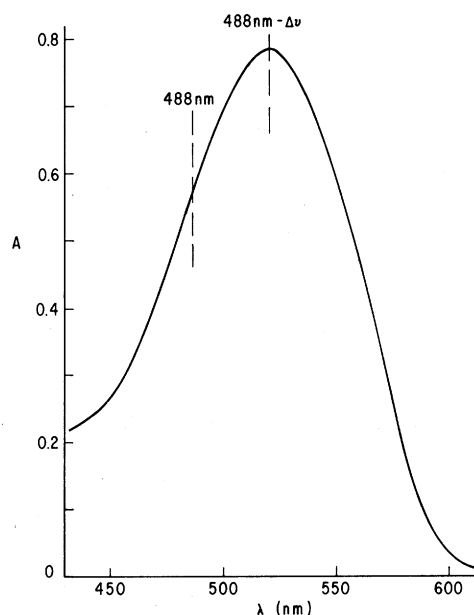


FIG. 2. Visible absorption spectrum of amaranth showing positions of incident and scattering wavelength. $\Delta\nu = 1347$ cm^{-1} , concentration $3.31 \cdot 10^{-5}$ M, pathlength, 1.00 cm.

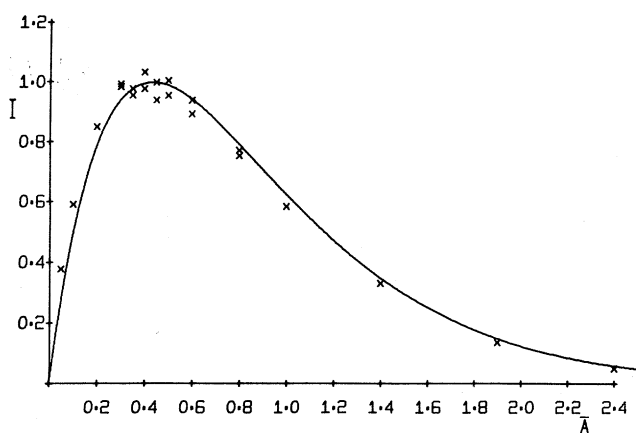


FIG. 3. Observed Raman intensity of the 1347 cm^{-1} band of amaranth in arbitrary units vs solution absorbance, averaged for incident and scattered wavelength for 1 cm pathlength. Solid curve calculated by Eq. (5) by a least squares adjustment of the combined constants I_0J .

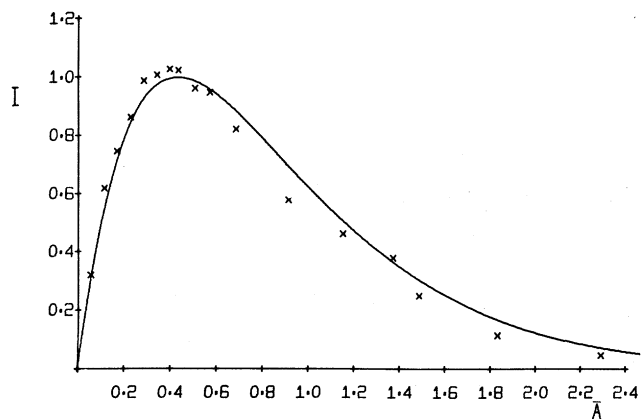


FIG. 4. Observed Raman intensity of the 1460 cm^{-1} band of tris(1,10-phenanthroline)iron(II) in arbitrary units vs solution absorbance, averaged for incident and scattered wavelength for 1 cm pathlength. Solid curve calculated by Eq. (5) by a least squares adjustment of the combined constants I_0J .

It is convenient to define an average absorbance for unit pathlength of the sample:

$$\bar{A} = c(\epsilon_s + \epsilon_i)/2 \quad (4)$$

Combining Eq. (4) with Eq. (1) and setting $r = l$ yields:

$$I = (2I_0J\bar{A}e^{-2kr\bar{A}})/(\epsilon_s + \epsilon_i) \quad (5)$$

Combining Eqs. (3) and (4) gives:

$$\bar{A}_{\text{opt}} = 1/(2kr) \quad (6)$$

Corresponding relationships can be derived for the general case,¹ $l \neq r$. Eq. (6) is particularly convenient because it does not involve absorptivity or concentration. Strekas *et al.* have applied Eq. (6) to the resonance Raman scattering of a tris(1,10-phenanthroline)iron(II) solution in a capillary cell of 0.1 cm diameter.¹ Using an experimentally determined \bar{A}_{opt} value of 13, they obtained an r value which is about one-third of the experimental value. Conversely, if the experimental r

value of 0.05 cm is used, \bar{A}_{opt} will be in error by a factor of 3. The authors suggested that the discrepancy reflects the inadequacy of the point-scattering model.¹ It is also conceivable that sample decomposition might have taken place in the capillary cell and that the geometry of such a cell is not well enough defined.

We have carried out similar measurements using the stable dye amaranth, trisodium salt of 3-hydroxy-4-[(4-sulfo-1-naphthalenyl)azo]-2,7 naphthalenedisulfonic acid. A Spex Industries Ramalog System* equipped with an argon ion laser was used for making the measurements. The laser power at the sample was ~ 300 mW. The resonance Raman spectrum of the compound is shown in Fig. 1 and the visible absorption spectrum in Fig. 2. A stationary Raman cell was employed, with $l = r = 0.5$ cm. A flow system was used to circulate the sample solution through the cell in order to avoid local overheating and sample decomposition. The strong Raman line at $\Delta\nu = 1347\text{ cm}^{-1}$ was used for intensity measurements; the 488 nm line of the argon ion laser was used for excitation.

The experimental points in Fig. 3 show measured values of I , the emerging scattered radiation, in arbitrary units as a function of \bar{A} . The solid line is calculated by Eq. (5) by a least squares adjustment of the constants I_0J . The experimental points follow the calculated curve in a satisfactory manner, indicating that in this case the point-scattering model is a valid approximation. It is evident that the experimental maximum is very close to the theoretical value, $\bar{A}_{\text{opt}} = [2kr]^{-1} = 0.434$, (if $r = 0.5$). If desired, c_{opt} can be calculated easily by Eq. (3).

We have repeated these measurements with tris(1,10-phenanthroline)iron(II), the dye used by Strekas *et al.*¹ Special precaution was taken to prevent decomposition and oxidation. Hydroxyl amine hydrochloride in potassium biphthalate buffer was added to prevent oxidation. Sample preparation was carried out in a nitrogen atmosphere. The sample solution was circulated through the Raman cell. The green 514.5 nm line of the laser was used for excitation. The I vs \bar{A} curve is shown in Fig. 4. Again the position of the intensity maximum agrees with the calculated value of 0.434 and the experimental points follow the calculated curve. We conclude that the point-scattering model as described by Strekas *et al.*¹ is a valid approximation if a larger Raman cell of known dimensions is used, and that Eqs. (2), (3), and (6) can be used to estimate the optimum concentration and the value of \bar{A}_{opt} under these conditions.

ACKNOWLEDGMENT

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1. T. C. Strekas, D. H. Adams, A. Packer, and T. G. Spiro, *Appl. Spectrosc.* 28, 324 (1974).

* The mention of commercial items is for your convenience and does not constitute an endorsement by the Department of Agriculture over other items of a similar nature not mentioned.